EXHIBIT 7

Response of German Cockroach (Orthoptera: Blattellidae) Populations to a Frequency Sweeping Ultrasound-Emitting Device

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ABSTRACT Populations of the field-collected *Blattella germanica* (L.) were confined in 1.2-m plywood cubes with food, water and an ultrasound-emitting device for 7 days. When pairs of cubes were connected by a length of tubing and one cube of the pair contained an active ultrasound-emitting device, significantly fewer cockroaches were found in the ultrasonic active cube. No significant change in cockroach distribution was noted in paired cubes with an inactive ultrasound-emitting device. Significantly more cockroaches were caught from cubes with an active ultrasound-emitting device when the tube was replaced by a bottle pitfall trap. The results of these experiments suggest that *B. germanica* activity was increased by the active ultrasound-emitting device.

THE USE OF ultrasonic sound (ultrasound) to eliminate or manage pests in the near environment has been widely publicized over the past 3 years. As interest and confusion in the use of ultrasound persists, public and private organizations have come under increased pressure to provide recommendations regarding this technology (Dunn 1982, Rambo 1982, Miller 1983).

Research on the use of ultrasound in agricultural and urban environments has been limited. Pulsed ultrasound was used with limited success to influence the behavior of various lepidopteran crop pests. Belton and Kempster (1962) reported a 63% reduction of the European corn borer, *Ostrinia nubilalis* (Hübner), in sweet corn. Payne and Shorey (1968) indicated a 31% reduction in oviposition in the cabbage looper, *Trichoplusia ni* (Hübner), in lettuce and broccoli. In contrast, Agee (1967) recorded no changes in populations of the cotton bollworm, *Heliothis zea* (Boddie), or the tobacco budworm, *H. virescens* (F.), in cotton; Agee and Webb (1969) reported no control of the cotton bollworm, *H. zea*, in cotton; Shorey et al. (1972) reported no control of the corn earworm, *H. zea*, in sweet corn in response to pulsed ultrasound. All researchers reported probable interference with sound shadows from the crop vegetation.

Adults of the cotton bollworm, *H. zea*, exhibited erratic flight patterns and evasive behavior when exposed to pulsed ultrasound both in caged and field experiments (Agee 1969, Agee and Webb 1969). However, in both situations, habituation and sound shadows were found to interfere with the moth's behavioral response to ultrasound. Resting, feeding, mating and oviposition continued with little interruption.

Laboratory experiments designed to measure the response of field-collected populations of the German cockroach, *Blattella germanica* (L.), to sound following exposure to one of seven sound frequencies ranging from 1 to 60 kHz have failed to influence the distribution of cockroaches in choice boxes (Ballard and Gold 1982, Ballard and Gold 1983a). Because of continuing requests for information on ultrasound, and because previous research was accomplished using steady-state pure tones emitted by a noncommercial device, our experiment was designed to reevaluate the response of field-collected German cockroach populations to a commercial, frequency-modulated ultrasound device.

Materials and Methods

The commercial ultrasound-emitting device used in this investigation was Pest Guard (manufactured by Global Innovators Inc., 17140 Norwalk Blvd., Suite 106, Cerritos, Calif.). Pest Guard is 14.5 by 8.5 by 7 cm in size and is designed for placement on horizontal surfaces. The device is powered by a standard 120-V AC, 60-Hz power source. According to literature provided by the manufacturer, Pest Guard sweeps through a frequency range of 30 to 65 kHz with an adjustable sweep rate of one to three sweeps per second.

Eight test chambers (cubes) were constructed of plywood with the finished surface to the inside of the cube (Fig. 1 and 2). Each cube had a removable lid and was 1.22 m on a side. Four cubes were assembled within each of two rooms. Each cube

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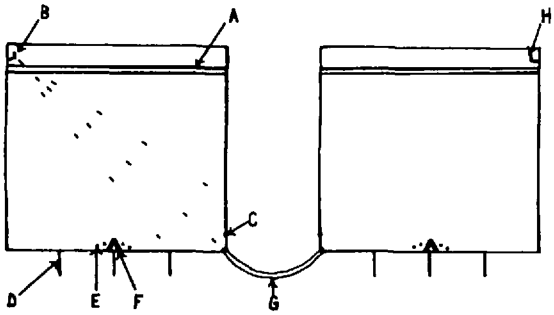

Fig. 1. Paired test cubes connected by plastic tube. (A) Electric shock barrier; (B) active ultrasound device; (C) sound pressure monitoring port; (D) support raising cube off floor; (E) Purina dog chow food; (F) water flask; (G) connection tube; (H) inactive (control) ultrasound device.

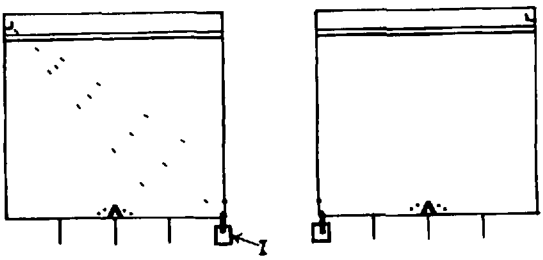

Fig. 2. Test cube connected to bottle trap. (I) Unbaited bottle trap fitted with 2-cm wide petroleum jelly barrier.

was raised off of the floor on 1.22-m lengths of pine (5 by 15 cm). The seams inside of each cube were caulked and all surfaces were painted with two coats of white enamel paint to minimize loss of sound through absorption in the walls and to maximize sound reflection. Maximizing sound reflection in a closed cube would result in a mixed or diffused sound where every point in the cube would have approximately equal sound (Krasil'nikov 1963). An interior electric shock barrier (Ballard and Gold 1983b) was located 15 cm below the top of the cube to prevent the escape of the cockroach population. The ultrasound device rested on a wooden shelf mounted in one top corner of the cube. The shelf was angled to direct the sound beam to the opposite lower corner of the cube (2 m distance). In that lower corner, a 4-cm hole was drilled and fitted with a rubber grommet for the attachment of either plastic tubing or a bottle pitfall trap. In one series of experiments, pairs of cubes were attached by a 90-cm length of clear plastic tubing (3 cm diam.). The tubing permitted cockroaches to move freely between cubes (Fig. 1). In another series of experiments the tube was removed and an unbaited pitfall trap (petroleum jelly barrier) was connected by a 10-cm vertical length of clear plastic tubing (3 cm diam.) (Fig. 2). Cockroaches could move freely between connected pairs of cubes or be caught in pitfall traps in unconnected tubes. Both procedures were used as a measure of the response of the cockroach population to ultrasound.

Field-collected German cockroach populations placed into the cubes consisted of 50 adult males, 50 adult females (no öotheca), and 200 nymphs. A 1:1:4 ratio was used because extensive field collections and trapping studies indicated that this ratio would best approximate the actual composition of a field population (Ballard and Gold 1983b). Seven replications with active devices and three replications with inactive devices were completed in cubes connected by tubes. In cubes fitted with pitfall traps, 10 replications were completed with both active and inactive ultrasound devices. Live cockroach counts were recorded once daily in each cube for the 7-day duration of each replication. Counting was done using a flashlight to minimize

Table 1. Characterization of the four Pest Guard ultrasound devices used in this investigation

Device	dB SPL At selected distances (m) in soundproof room[a]				dB SPL At selected sweep settings in soundproof room[b]				Sweeps/sec at selected sweep settings in soundproof room[b]				dB SPL At 2 m in test room[c]	$\bar{x}$ Sound pressure (dBSPL) in cubes containing active ultrasound device[d]
	0	1	2	4	2	4	6	8	2	4	6	8		
1	120.0	95.5	90.5	84.0	96.0	96.4	96.0	96.0	2.0	2.0	2.8	3.5	60.0	54.2
2	120.0	97.4	91.0	84.0	97.8	97.8	97.8	97.8	2.0	2.0	2.5	4.0	62.0	55.3
3	120.0	95.0	88.0	81.0	95.0	95.0	95.0	95.0	1.8	2.0	2.0	2.8	63.0	55.5
4	120.0	94.6	89.3	83.5	94.6	95.0	94.6	94.6	2.0	2.0	2.5	2.8	68.5	58.8

[a] Bruel and Kjaer Type 2209 sound level meter fitted with a Bruel and Kjaer Type 4138 microphone (0.4 cm diam.) and a Bruel and Kjaer Type 1616, 1/3 octave filter set with a 0.0002 dyne/cm^2 standard.
[b] Number of times the device sweeps through the 30 to 65 kHz frequency range per second.
[c] Bruel and Kjaer Type 2203 portable precision sound level meter fitted with a microphone (2.54 cm diam.) and Type 1613 octave filter set with a 0.0002 dyne/cm^2 standard.
[d] Mean sound pressure measured with portable sound level meter in cube containing an inactive ultrasound device was ca. 12 dB SPL, which is the same as the internal noise of the sound level meter measured in a soundproof room.

Table 2. Mean daily number of German cockroaches in each cube

Day	Inactive	In tube[a]	Inactive[b]	Inactive	In tube[a]	Active[c]
1	154.3	68.0	151.3	188.1	45.9	170.6
2	132.3	87.0	125.7	157.0	58.0	134.1
3	100.0	83.0	88.0	143.1	66.6	109.9
4	80.3		66.7	119.7	99.0	89.7
5	63.3		61.0	108.4		86.6
6	49.3		66.7	97.0		70.3
7				81.0		76.8

[a] Control cube ultrasound (inactive device) was connected to another control cube by plastic tubing. Three replications, 300 cockroaches per cube.
[b] A cube containing an active ultrasound device was connected to another cube containing an inactive ultrasound device. Seven replications, 300 cockroaches per cube.
[c] So many cockroaches were found in the connecting tube that they could no longer be counted beyond the first 3 or 4 days.

disturbance of the cockroaches. In addition, the cockroaches in either the connecting tube or in each pitfall trap were counted.

The local distributors of Pest Guard provided eight devices for the research program. Four devices, numbered one through four, were characterized acoustically and used in all active device replications. The other four devices were always used as inactive controls. Characterization of the four devices was accomplished in a sound-isolated room through the use of a nonportable Bruel and Kjaer Type 2209 sound level meter fitted with a 0.4-cm-diameter Bruel and Kjaer Type 4138 microphone and a Bruel and Kjaer Type 1616, ⅓ octave filter. The influence of the adjustable sweeps per second capability of the device was evaluated with a digital oscilloscope, which also printed a tracing of the number of sweeps per second. The sound pressure within each cube (whether with an active or inactive device) was measured with a portable Bruel and Kjaer Type 2203 precision sound level meter fitted with a Type 1613 octave filter. The microphone (2.54 cm diam.) was inserted into the port drilled in the cube just above the exit hole (Fig. 1). Analysis of variance and simple linear regression were used to analyze the data; means were separated by Duncan's (1951) multiple range test.

Results and Discussion

The four active ultrasound devices (Table 1) revealed no significant differences in their performance (decibels sound pressure level [dB SPL]) at the selected distances and sweep settings tested. All evaluations in the cube were based on sweep setting number four. This was the only setting which produced a uniform number of sweeps per second (two) among the four devices tested. The considerable difference in sensitivity (ca. 30 dB SPL) between the laboratory and the portable sound level equipment was due to the difference in microphones in these devices. Because dB SPL measurement is a logarithmic relationship, the sound within the cube was considerably more intense than was measured by the portable sound level meter.

The Pest Guard devices tested swept through frequencies of 30 to 65 kHz from 1.8 to 4 times per second (Table 1). Because no appreciable changes in dB SPL were noted at the selected sweep settings, the sound energy emitted by these devices was in the form of an FM ramp, rather than of osculating pulses. The intensity of sound pulses depends upon environmental conditions.

When the influence of ultrasound was evaluated with paired cubes connected by plastic tubing, significantly fewer cockroaches ($P \leq 0.10$) were found

Table 3. Mean daily catch as percentage of live German cockroach population remaining in cubes for 7 days

Day	Inactive[a]	Active[b]
1	15.54a[c]	24.63a
2	15.91a	24.69a
3	15.80a	24.97a
4	17.01a	19.71abc
5	15.00a	22.49ab
6	12.43a	14.57c
7	9.97a	16.23bc

Cockroaches caught in a bottle pitfall trap fitted to exit hole.
[a] Control cube contained an inactive ultrasound device and a population of 300 cockroaches, 10 replications.
[b] Active cube contained an active ultrasound device and a population of 300 cockroaches, 10 replications.
[c] Means within a column followed by the same letter were not significantly different ($P \leq 0.10$; Duncan's [1951] multiple range test).

Table 4. Mean number of German cockroaches alive per day in cubes fitted with pitfall bottle trap

Day	Inactive[a]	Active[a]
0	300.0	300.0
1	232.7	239.2
2	230.1	237.8
3	224.3	235.3
4	213.9	233.3
5	213.3	226.8
6	205.4	223.2
7	203.5	222.2

Populations of 300 cockroaches in each of 10 replications. The number of cockroaches alive per day was the number of living cockroaches counted in the cube plus the cumulative number of cockroaches caught in the trap each night.
[a] No significant difference in mortality (ANOVA). Considerable mortality noted in cubes the day before initiation of test due to handling of field-collected cockroaches.

in the cubes containing an active ultrasound device than in the connected cubes with an inactive device (Table 2). There was no significant difference in the distribution of cockroaches within the control pairs of cubes. Significantly more cockroaches ($P \leq 0.10$) were found in the tube connecting the control cubes than in the tube connecting the active and inactive cubes. That large numbers of cockroaches were found in the connecting tube whether ultrasound was present or not was an undesirable design feature. Although the results of this experiment indicated that cockroaches responded to ultrasound under these test conditions, the experiment was repeated with an improved design.

To improve the experimental design, the connecting tubes were removed and the cubes fitted with a pitfall trap. The analysis of data indicated a significant increase ($P \leq 0.10$) in the rate of cockroach catch in the cubes with an active ultrasound device (Table 3). Ultrasound caused an increase in movement in the confined cockroach population, which resulted in a significant increase in cockroach catch. Statistical separation of catch means on a daily basis indicated habituation in days 6 and 7. When the daily cockroach catch means of both control and active cubes were combined and statistically compared, the catch rates on days 6 and 7 in the active cube were not significantly different from the catch rates in the control cube.

Although the field-collected cockroaches had considerable natural mortality, there was no significant difference in daily mortality between control and active cubes (Table 4). These results were consistent with the advertised claims that Pest Guard does not kill pests.

The use of ultrasound in these experiments resulted in a statistically significant increase ($P \leq 0.10$) in activity by the German cockroach population. However, the biological importance of these observations is difficult to interpret. Probably, German cockroaches randomly move from exposed areas to sound shadows to escape ultrasound. In the home environment, perceived reductions in cockroach populations may satisfy the aesthetic threshold, but the same number of cockroaches could still exist in the structure but in different locations. Whether or not control is achieved remains to be evaluated.

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